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# Influence of different factors on the stability of biodiesel: A review

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ABSTRACT

Gradual depletion of fossil fuel has greatly enhanced the necessity to look for alternative fuel for automotive engine. In response to this, biodiesel is being considered as a promising solution with a number of technical advantages over conventional petroleum diesel. However, commercial use of biodiesel has been limited because of some drawbacks including corrosivity, instability of fuel properties, higher viscosity, etc. Stability of fuel properties is especially important to ensure expected engine performance as well as engine life. Most of the research papers available in public domain reported some concerns on the stability of biodiesel fuel properties. Scattered studies on different aspects often lead to confusions in understanding the adverse effects of different factors on the degradation of biodiesel. In this article, different factors that cause instability in biodiesel and their possible implications of different fuel properties have been reviewed. A special emphasis has been given to explain the possible mechanisms of biodiesel degradation. Possible remedies to improve the stability of biodiesel have also been included. Finally, some general conclusions concerning these problems are summarized and further scopes of study have been suggested.



**Contents**



## 1. Introduction

Energy has become an indispensable factor for mankind to preserve economic growth and maintain living standard [1]. In recent years, consumption of energy is increasing rapidly due to tremendous growth of transportations and industrial sectors [2].Therefore, considerable attention has been given to look for alternative fuels. Biodiesel is being considered as one of the most promising alternative fuels [3]. It is nontoxic in nature, biodegradable and produced from renewable sources such as vegetable oils or animal fats [4,5]. Biodiesel is completely miscible with diesel and thereby can also improve certain fuel qualities [6].

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For instance, addition of biodiesel in diesel can improve lubricity as inherently biodiesel has better lubricity. Biodiesel having 11% oxygen by weight [7,8] can ensure complete combustion and thereby increases the performance of engine. It also reduces the emission of exhaust gases as compared to that of diesel [9]. However, the commercial use of biodiesel has been limited due to having some drawbacks. Biodiesel has higher viscosity which may result in injector coking, moving parts sticking, fuel filter plugging, etc. [10]. Higher viscosity also causes poor atomization and less accurate operation of fuel injectors [11]. What makes the situation more complete is the fact that biodiesel is auto-oxidative and can readily influence its other fuel properties including cetane number, flash point, viscosity, density and so on.

There are many factors, which can influence the stability of biodiesel. It has been reported that biodiesel is highly sensitive to light, temperature and metal ions [12], more hygroscopic in nature [13], more susceptible to oxidation reactions [14,15] and more corrosive than diesel [16–20], unless it is modified or treated with additives [21]. Degradation of biodiesel causes its compositional changes [17–19], and consequently it changes its different valuable fuel properties [22]. Degradation of biodiesel also has strong influence on the corrosion of metals [13] as well as tribological aspects [19,23]. Therefore, it is very important to understand the influence of different factors on the instability of biodiesel and their related mechanisms. Understanding of such things is expected to contribute for improving stability of biodiesel [24].

From the recent literature studies, it has been found that the properties of biodiesel could be deteriorated by several factors including (i) auto-oxidation, (ii) thermal decomposition or thermal fluctuations, (iii) water absorption, (iv) biodegradation with microbial growth, (v) storage conditions, (vi) metal contamination, (vii) presence and absence of additives, etc. The available scattered studies do not allow understanding the real facts causing instability of biodiesel. The present study has critically analyzed these factors and come out with the possible mechanisms of instability of biodiesel.

## 2. Biodiesel and its specification

Globally, there are more than 350 oil-bearing crops identified as potential sources for biodiesel production [25]. The most common of them are soybean oil [26,27], sunflower, corn, used fried and olive oil [28], rapeseed oil [29–31], castor, lesquerella oil [32], milkweed (Asclepias) seed oil [33], *Jatropha curcas* [34], *Pongamia glabra* (Karanja), *Madhuca indica* (Mahua) and *Salvadora oleoides* [35], palm oil [36–38], etc. In general, biodiesel derived from these sources can be defined as mono-alkyl esters of long chain fatty acids [39]. Vegetable oils/animal fats mainly consist of triglyceride molecules. Triglycerides in the presence of catalyst react with methanol and thereby reduce methyl esters and glycerol (Fig. 1).

Glycerol is removed as by-product. The esters produced are known as biodiesel. According to ASTM D6751, standard biodiesel should have minimum 96.4% esters. The rest could be water, sediment, free glycerol, methanol, monoglyceride, diglyceride, triglyceride, alkaline metals (Na+K), alkaline earth metals (Ca+Mg), etc. The presence of these components can greatly influence the properties of fuel. As per standard, the limit of these contaminants and the level of desired fuel properties are shown in Table 1. Although the properties of biodiesel meet the given standard in as-receive condition, many properties can deviate from the standard once biodiesel is degraded. Therefore, the factors that may cause instability of biodiesel in terms of its composition as well as related fuel properties are very important to study in systematic way. In response to this, the following section has been devoted to describe the significance of fuel stability and the factors that may make it instable.

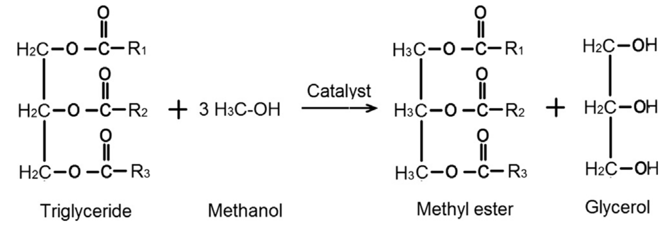



**Fig. 1.** Chemistry of transesterification reaction ($R_1$, $R_2$, and $R_3$ represent the hydrocarbon chains of the fatty acid triglyceride) [40].

## 3. Instability of biodiesel

Instability of biodiesel refers to the change of its original fuel composition and fuel properties. In such case, biodiesel properties can deviate from the prescribed quality standard. This can greatly affect its applicability in automotive engine. Engine performance as well as fuel consumption rate is greatly altered due to deterioration of fuel [40]. As compared to diesel, biodiesel is much more instable [41]. Therefore, manufacturers are concerned to extend the warranty of automotive while biodiesel/its blend with diesel are used as fuel.

Biodiesel is composed of different saturated and unsaturated fatty acid esters. The level of saturated and unsaturated components depends on the type of biodiesel feedstock. Fig. 2 shows the composition of different biodiesels. Unsaturated components in biodiesel chemically are more reactive. Based on the percentage of unsaturated components shown in Fig. 2, it can be said that palm and coconut biodiesel could be chemically more stable compared to others. In addition, the presence of oxygen makes it more aggressive to react with metal surface. The hygroscopic nature of biodiesel also leads to the formation of considerable amount of microorganism which alters its local chemistry [42]. Any chemical reactions occurring in biodiesel can change its fuel composition. It has been reported that biodiesel, being degraded can form different sort of acids, aldehyde, ketone, insoluble sediments etc. Formation of these compounds can greatly change its fuel properties including total acid number, viscosity, density, flash point, cetane number, etc. Degradation of these properties is directly related to engine performance and durability. For example, increased viscosity can cause fuel operation and atomization problem [2], thus increasing the tendency for coking of injector, sticking of moving parts, plugging of fuel filter, etc. Because of its instability, biodiesel is not yet being considered for commercial use in automobile engine. Interaction between biodiesel and engine oil is another concern in engine operation. During any engine operation, fuel will always penetrate into the sump (the lower part of the crankcase of an internal-combustion engine into which fuel can drain form a reservoir). The penetration of fuel is known as fuel dilution. Fuel dilution is more pronounced for in-cylinder diesel particulate filter (DPF) regeneration as post-injection of fuel to raise exhaust gas temperature in cylinder and burning off carbon-based deposits blocking diesel particulate filter (DPF) [6,43]. Biodiesel has lower volatility and causes sedimentation in the sump. As the blend ratio of biodiesel increased more with petroleum diesel in-cylinder, DPF regeneration can lead to higher fuel dilution [6]. In addition, oxidation instability of biodiesel leads to higher fuel dilution and increased viscosity of engine oil. Therefore, it is very important to study on the biodiesel stability influencing factors and necessary remedial measures to use it as an efficient and compatible diesel engine fuel.

**Table 1**
Specification of biodiesel as per ASTM D6751 and EN 14214 standards (ASTM, 2008; EN, 2008) [41–43].

| Property | Test methods and limits | | | |
|---|---|---|---|---|
| | ASTM D6751 | Limits | EN 14214 | Limits |
| Density at 15 °C (kg/m$^3$) | – | – | EN ISO 3675 | 860–900 |
| Kinematic viscosity at 15 °C (mm$^2$/s) | D445 | 1.9–6.0 | EN ISO 3104 | 3.5–5.0 |
| Cetane number (min) | D613 | 47 | EN ISO 5165 | 51 |
| Flash point (°C) | D93 | 93 | EN ISO 3679 | 101.0 |
| Copper strip corrosion | D130 | No. 3 max | EN ISO 2160 | Class 1 |
| Cloud point (°C) | D2500 | Report | – | – |
| Oxidation stability at 110 °C (h) | – | – | EN 14112 | 6 |
| Acid value (mg KOH/g) | D664 | 0.50 max | EN 14104 | 0.5 max |
| Iodine value | – | – | EN 14111 | 120 max |
| Polyunsaturated (P4 double bonds) | – | – | EN 14103 | 1 max |
| Water and sediment (vol%) | D2709 | 0.050 max | – | – |
| Water content (mg/kg) | – | – | EN ISO 12937 | 500 max |
| Ester content % (m/m) | – | – | EN 14103 | 96.5 min |
| Free glycerol % (m/m) | D6584 | 0.020 max | EN 14105 | 0.02 |
| Total glycerol % (m/m) | D6584 | 0.240 max | EN 14105 | 0.25 max |
| Methanol content % (m/m) | – | – | EN 14110 | 0.2 max |
| Monoglyceride content % (m/m) | – | – | EN 14105 | 0.8 max |
| Diglyceride content % (m/m) | – | – | EN 14105 | 0.2 max |
| Triglyceride content % (m/m) | – | – | EN 14105 | 0.2 max |
| Alkaline metals (Na+K) (mg/kg) | – | – | EN 14108 | 5 max |
| Alkaline earth metals (Ca+Mg) | – | – | EN 14538 | 5 max |
| Total contamination (mg/kg) | – | – | EN 12662 | 24 max |

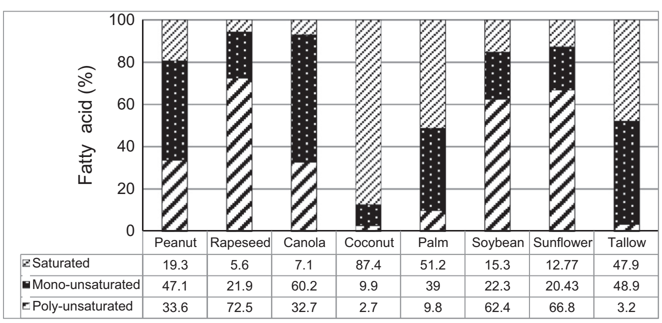



**Fig. 2.** Saturated, mono- and poly-unsaturated fatty acid contents for different biodiesel [47].

## 4. Factors affecting stability

### 4.1. Auto-oxidation

Biodiesel having unsaturated fatty acid components along with more carbon–carbon double bonds and fewer hydrogen molecules is more susceptible to oxidation. When biodiesel is exposed to oxygen, the oxygen itself attaches to the bis-allylic site directly adjacent to the double bonds. This initiates the auto-oxidation chain reactions and ultimately forms peroxides. The process of peroxidation promotes auto-oxidation by three steps: initiation, propagation, and termination [44]. The first set involves the removal of hydrogen from a carbon atom to produce a carbon free radical [45]. Secondly, the new carbon free radical then reacts with diatomic oxygen to continue the propagation cycle. Thirdly, the chain reaction terminates when two free radicals react with each other to yield stable products. The oxidation can be presented as follows [44]:

$$\text{Initiation}: RH + I^{\bullet} \rightarrow R^{\bullet} + IH \ (I^{\bullet} = \text{initiator radical})$$

$$\text{Propagation}: R^{\bullet} + O_2 \rightarrow ROO^{\bullet} \ (ROO^{\bullet} = \text{peroxide radical})$$
$$ROO^{\bullet} \rightarrow ROOH + R^{\bullet}$$

$$\text{Termination}: R^{\bullet} + R^{\bullet} \rightarrow R-R$$
$$ROO^{\bullet} + ROO^{\bullet} \rightarrow \text{stable product}$$

In auto-oxidation reactions, normally oxygen free radicals ($HO^{\bullet}$, $ROO^{\bullet}$, $HOO^{\bullet}$, $O^{\bullet}$ and similar) are generated radiolytically. These free radicals will readily abstract a hydrogen atom from the hydrocarbon chain and thereby lead to polymerization. However, this auto-oxidation process depends on many variables such as temperature, light radiation intensity, presence of naturally occurring antioxidants, degree of unsaturated molecules, oxygenated moieties, metallic traces and more [12,14,46]. However, from the above reactions, it seems that the presence of unsaturated ester molecules as well as oxygenated moieties is the major cause of auto-oxidation. Auto-oxidation of biodiesel can cause degradation of fuel quality by forming oxidation products such as aldehydes, alcohols, shorter chain carboxylic acids, insoluble gums, sediments, etc. [47]. These oxidation products can reduce engine life by introducing different sort of problems. The most common problems could be fuel filter plugging, injector fouling, deposit formation as well as wear, corrosion and tribo-corrosion of fuel components [48]. The oxidation stability can be increased by adding additional natural and synthetic antioxidants. Related mechanism of anti-oxidation has been explained later.

### 4.2. Storage stability

Storage stability is concerned when biodiesel is stored for longer time. Compositional change is probably one of the primary concerns of storage stability. In addition, storage stability could be influenced due to exposure to air [49,50], sunlight, exposed metal surfaces, sometimes changing the storage container [51], temperature [52], etc. It has been reported that because of losing stability, biodiesel after 6-month storage cannot be used as fuel. Several researchers have reported the effect of storage stability on the properties of biodiesel and its composition. Most of the investigated fuel properties include acid value (AV), peroxide value (PV), induction period (IP), flash point, iodine value (IV), density, viscosity, etc. The significance of these properties is listed in Table 2. The effect of storage stability on these fuel properties reported by several researchers has been summarized in Table 3.

From Table 3, it is noted that certain properties of biodiesel such as peroxide value (PV), acid value (AV), density (D) and viscosity (V) increase with the increase of storage time.

**Table 2**
General properties for measuring the stability of biodiesel.

| Properties of biodiesel | Indication criterion | Ref. |
|---|---|---|
| Induction period (IP) | Indicate the initiation of oxidation by measuring the conductivity of fuel. Lower the IP lower the oxidation stability. | [57] |
| Acid value (AV) | The peroxide formed during oxidation finally turned into organic acids, which is determined by AV. Higher the acid value, higher the formation of acidic products by oxidation. | [58] |
| Peroxide value (PV) | Determines the oxidation progression and indicates the formation of secondary oxidation products. Higher PV values, higher the rate of peroxide formation. | [59] |
| Iodine value (IV) | IV measures the level of unsaturation in organic compounds, such as FAME (fatty acid methyl ester). Lower iodine number is resulted due to the lower degree of unsaturation. | [59] |
| Density (D) & viscosity (V) | Oxidation products are higher in molecular weight and thus increase density, which also results in higher viscosity. | [60] |
| FAME concentration | FAME concentration determines the composition of biodiesel, which indicates the degradation of fuel. | [54] |

**Table 3**
Effect of storage stability of different biodiesels on different fuel properties (↑ Increase in quantity; ↓ Decrease in quantity).

| Types of biodiesel | Storage periods | Measured properties and relative changes | Storage limit based on stability of properties | Ref. |
|---|---|---|---|---|
| High oleic sunflower oil | 30 months | PV ↑, IV ↓, AV ↑, V ↑ | 12 months | [61] |
| High erucic Brassica oil | 30 months | PV ↑, IV ↓, AV ↑, V ↑ | 12 months | [61] |
| Low erucic Brassica oil | 30 months | PV ↑, IV ↓, AV ↑, V ↑ | 12 months | [61] |
| Karanja oil | 6 months | IP ↓ | 4 months | [62] |
| Rice bran oil | 24 months | PV ↑, IV↓, AV ↑, V ↑ | 1 month | [63] |
| Rapeseed oil | 200 days | PV ↑, IP ↓, V ↑ | 60 days | [53] |
| Used frying oil | 200 days | PV ↑, IP ↓, V ↑ | 50 days | [53] |
| Palm methyl ester | 3 months | AV ↑, V ↑, IP ↓ | 1 month | [64] |
| Jatropha methyl ester | 3 months | AV ↑, V ↑, IP ↓ | 1 month | [64] |
| Coconut biodiesel | 3 months | AV ↑, V ↑, IP ↓ | 1 month | [64] |
| Soybean biodiesel | 3.5 years | IP ↓ | 14 weeks | [65] |
| Canola oil | 190 days | FAME concentration ↓ | 15 days | [54] |
| Linseed oil | 60 days | IP ↓ | 40 days | [66] |
| Lard oil | 60 days | IP ↓ | 30 days | [60] |
| Tallow oil | 60 days | IP ↓ | 30 days | [60] |
| Neat edible rapeseed oil | 52 weeks | Purity of Biodiesel ↓ | 20 weeks | [67] |
| Sunflower oil methyl ester | 12 months | IP ↓ | 6 months | [68] |
| Non distilled Karanja biodiesel | 12 months | PV ↑, AV ↑, V ↑ | 4 months | [55] |
| Soybean (edilbe oil) | 10 months | IP ↓ | 6 months | [69] |
| Sunflower oil ethyl ester | 40 days | IP ↓ | – | [70] |
| Mahua oil methyl ester | 12 months | PV ↑, V ↑ | – | [71] |

*Note*: peroxide value (PV), iodine value (IV), acid value (AV), viscosity (V), density (D), induction period (IP).

Therefore, it can be concluded that the newly generated biodiesel components upon degradation due to long-term storage are acidic in nature and always will have higher density and viscosity as well. As per prescribed limit in standard, the acceptable storage duration of different biodiesels could vary from 1 to 12 months. Further study should be done to clarify this finding. It is also noticeable from Table 4 that the iodine value/degree of unsaturation and induction period decreases with the increase of storage time. This clearly demonstrates that compositional change or degradation of biodiesel is directly related to degree of unsaturated molecules.

### 4.3. Thermal decomposition

Temperature plays an important role on the deterioration of the fuel quality. The tendency of a fuel exposed to elevated temperature for forming asphaltenes is a representation of thermal stability. Asphaltenes produced in high temperature can plug the fuel filters of the engines because generally asphaltenes act as resinous substances like tar [41]. Due to thermal decomposition, biodiesel can produce different sort of products. For example, fatty acid methyl ester (FAME) of biodiesel starts isomerizing to more stable conjugated structure at high temperature when it is associated with methylene-interrupted polyunsaturated olefin structure [53,54]. After this isomerization, a cyclohexene ring is formed when adiene group (conjugated) from one fatty acid chain reacts with oneolefinic group from another fatty acid chain [54,55]. The reaction is titled as "Diels Alder reaction" as shown in Fig. 3. This normally occurs at 250–300 °C or above and the products of the reaction are mostly dimmers [56,57].

Many properties of biodiesel are greatly influenced by temperature. Those include oxidation, lubricity, corrosivity, composition, induction period, viscosity, density, etc. Stragevitch et al. [58] analyzed molecular structure of species involved in the oxidation reactions of unsaturated fatty acid esters at different temperatures. Among all the esters studied, ricinoleate was found to be the least susceptible to oxidation at elevated temperature. Several studies [59] have found that increasing temperature enhances respiration. Respiration is the rate of formation of $CO_2$ in the presence of $O_2$ that causes the degradation of fuel. Xin et al. [60] investigated the effect of temperature on the particular composition of biodiesel such as tocopherol. They observed that the remaining tocopherol (unreacted) decreased with the increase of temperature above 300 °C. Fazal et al. [61] reported that increase of temperature can enhance the oxygenated molecules and thereby improves the lubricity of diesel fuel. It has also been reported [62] that oxidation at elevated temperature may produce different products such as aldehydes, ketones, carboxylic acids, etc.

Lapuerta et al. [63] investigated the stability of soybean and cooking oil biodiesel for assessing the effect of different temperatures. It was evident that induction period decreases non-linearly when the temperature is increased. Nzikou et al. [64] investigated the thermal stability of biodiesels derived from vegetable oils. They observed that viscosity of soybean oil and methlyllic oil biodiesel increased with frying temperature from 20 °C to 180 °C.

**Table 4**
Effect of metal contamination on different biodiesels (↑ Increase in quantity; ↓ Decrease in quantity).

| Types of biodiesel | Physical states of metals exposed | Measured properties and changes | Relative influence of metals in degradation | Ref. |
|---|---|---|---|---|
| Jatropha biodiesel | Metal napthenates | IP ↓ | Cu > Co > Mn > Ni > Fe | [89] |
| Jatropha Curcas | Metallic powder | IP ↓ | Cu > Co > Mn > Ni > Fe | [95] |
| Palm biodiesel (B100) | Metallic coupons | AV ↑ | Cu > leaded bronze | [50] |
| Palm biodiesel (B50) | Metallic coupons | AV ↑ | Cu > leaded bronze | [50] |
| Palm biodiesel (B100) | Metallic coupons | Composition ↓ | Cu | [17] |
| Palm biodiesel | Metallic powders | IP ↓ | Cu > Co > Mn > Ni > Fe | [96] |
| Neem biodiesel | Metallic container | Metallic traces ↑ | – | [97] |
| Soybean biodiesel | Metallic sheets | Composition ↓ | Carbon steel | [98] |
| Sunflower biodiesel | Metallic sheets | Composition ↓ | Carbon steel | [98] |
| Palm biodiesel (B100) | Metallic coupons | Water content ↑, AV ↑ | Mild steel | [15] |
| Palm biodiesel (B50) | Metallic coupons | Water Content ↑, AV ↑ | Mild steel | [15] |
| Palm biodiesel (B100) | Metallic coupons | Composition ↓, AV ↑ | Cast iron | [21] |
| Soybean biodiesel | Metallic coupons | IP ↓, AV ↑ | Carbon steel > galvanized steel | [99] |
| Animal fat derived biodiesel | Metallic containers | PV ↑, IP ↓ | Cu > Ni > Fe | [59] |
| Palm biodiesel | Metallic Sheets | IP ↓ | Cu > Sb > Zn > aluminum alloys > Al > Fe | [100] |
| Palm biodiesel | Metallic coupons | Composition ↓, AV ↑ | Cu > Al > stainless steel | [19] |

*Note*: peroxide value (PV), iodine value (IV), acid value (AV), viscosity (V), density (D), induction period (IP).

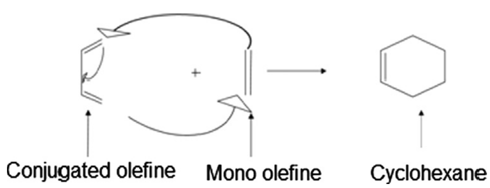



**Fig. 3.** Diels Alder reaction for thermal degradation of biodiesel [77].

Fernandes et al. [65] found that thermal stability of cottonseed oil was higher than that of cottonseed biodiesels. Albuquerque et al. [66] found that there was a decreasing trend of the oxidation temperature (OT) with the increase of bis-allylichydrogen (H) in the mixtures for all biodiesel samples with increasing temperature. These demonstrate that variation of composition is the key factor for the oxidation stability of biodiesel.

Garcia et al. [67] investigated thermal stability of Brazilian Cerrado by TGA analysis. They found that all biodiesel samples began to degrade after 130 °C and thus limiting the industrial use of those biodiesels. Dunn et al. [68] experimented methyl soyate (SME) to evaluate the accelerated oxidation under varying time and temperature and found that with increase in reaction temperature, acid value, specific gravity, temperature peroxide value, and viscosity increased but the cold flow properties were changed a little up to150 °C. The research works done so far lead to the conclusion that temperature has a great influence in changing composition as well as other fuel properties. This could vary depending on the types of biodiesel and its compositional or molecular structures.

### 4.4. Metal contamination

Metals can perform as a catalyst for oxidation and degradation of biodiesel [69,70]. Trace metals play a significant role in clogging vehicle fuel lines and can leave an undesirable residuum of metal oxides in engine parts. The presence of metallic elements is undesirable, even in low concentrations to control the pollution due to their release into the atmosphere through combustion of fuel [22,71]. At the initial step of biodiesel oxidation, the peroxides degrade by a number of pathways to secondary oxidation products, such as acids, aldehydes, dimers and polymers [72]. The initiator radical for peroxide formation can be formed by metallic ions as represented by the following [72]:

$$\text{Initiation by metallic ions:}\quad RH + I^{\bullet} = R^{\bullet} + IH$$
$$\text{Propagation by metallic ions:}\quad ROOH + M^{2+} = RO^{\bullet} + OH + M^{3+}$$
$$ROOH + M^{3+} = ROO^{\bullet} + H^{+} + M^{2+}$$

This peroxide radical ($ROO^{\bullet}$) reacts further as stated earlier to promote the oxidation process.

Several studies have been done to understand the effect of metal contamination on biodiesel stability. Metal assisted oxidation results in the formation of insolubles, gums and sediments along with corrosion products. Most of the investigations include metal exposure to biodiesel in static condition [69] sometimes with temperature fluctuations [13,73], time variance or with antioxidants [74]. In addition, some studies include dynamic situations [73]. The relative effect of several metals was measured by determining the change in biodiesel properties such as IP, AV, PV, Compositional analysis, water content, metallic traces and analysis of corrosion products. The studies conducted on metal exposed biodiesel are shortlisted in Table 4 with the measured changes in biodiesel properties.

From Table 4, it is seen that due to metal exposure, induction period is reduced while acid value (AV), peroxide value (PV), and water content are increased. Degradation of these properties has been observed to a greater extent due to exposure of copper as compared to other metals. Poor stability linked to metal contamination can be a great issue for commercial use of biodiesel. Therefore, careful choice of metal selection has a great importance in case of biodiesel.

### 4.5. Water absorption

The quality of biodiesel fuel is significant for its successful use in diesel engines and its subsequent replacement of nonrenewable fossil fuels. The presence of water in biodiesel has a strong effect on its fuel properties. The water content of biodiesel must be below 0.05% according to ASTM D6751. Biodiesel is hygroscopic in nature and therefore the content of water is a major issue of concern. Water can cause degradation of esters through hydrolysis and thereby change the fuel composition and property as well.

$$\underset{\text{(esters)}}{R_1-COOR_2} + \underset{\text{(water)}}{H_2O} \rightarrow \underset{\text{(alcohol)}}{R_1-OH} + \underset{\text{(fatty acid)}}{R_2-COOH} \quad \text{(hydrolysis of esters)}$$

The presence of water can also enhance the microbial growth, which may cause corrosion of metal surfaces. There are several number of studies related to degradation of biodiesel. DeMello et al. [75] observed that the rates of hydrolysis are slower than the microbial degradation rates of biodiesels. However, abiotic hydrolysis could become more relevant in conditions where microbial degradation is less ideal. Burton [76] has found that biodiesel can absorb 150 ppm of moisture where petro-diesel can only absorb 50 ppm of moisture.

According to Demirbas [77] the moisture absorption of biodiesel sufficiently decreases the heat of combustion. It may begin to form ice crystals when the temperature is near 0°. These crystals result in the formation of sites for nucleation and enhances the gelling of the fuel residue. It has been reported that water works as a part of the respiration system for most microbes. In addition, colonization of microbe and formation of gel can cause different technical problems (e.g. fuel system plugging, injector coking, etc.). Leung et al. [52] found that moisture content in biodiesel enhanced biodiesel degradation because of hydrolysis but it lessens to a great extent when the fuel is exposed to air and high temperature. Waste soybean derived biodiesel suffered hydrolytic degradation because of the cooking process and that destroys natural antioxidants of the oil [78]. Srivastava and Prasad [79] observed that biodiesel containing high content of intermediate products (mono- and diglycerides) will have the tendency to absorb water to a higher degree than biodiesel constituting mainly methyl ester. Felizardo et al. [80] found that fuel contaminated with water can cause engine corrosion. According to Fazal et al. [19] enhanced corrosiveness of biodiesel occurs due to higher moisture absorption, presence of oxygen moieties, and fatty acids produced from auto-oxidation. Crookes [81] noted that to reduce the further emissions, fuel emulsification with water can be used. Sharma et al. [82] reported that natural antioxidants contained in biodiesel are destroyed during the purification step thus making biodiesel susceptible to degradation. Therefore, it is very important to find suitable additive for reducing the absorption of moisture in biodiesel.

### *4.6. Microbial growth*

One of the major problems for the commercial use of biodiesel or its blend with diesel is biodegradation which causes changes of fuel properties [83]. Microorganisms in biodiesel have potential influence to accelerate the degradation process [84]. This section presents an updated overview on comparative degradation of petroleum diesel and biodiesel by microorganisms.

Petroleum diesel is composed of different hydrocarbons. It has been reported that hydrocarbons are biodegraded primarily by organic pollutants such as bacteria, yeast, and fungi. According to Adebusoye et al. [85] the microbial degradation of petroleum hydrocarbons can be occurred by several types of bacteria including *Pseudomonas fluorescens*, Bacillus sp., *Bacillus subtilis*, Alcaligenes sp., *P. aeruginosa*, *Acinetobacter lwoffi*, Flavobacterium sp., *Micrococcus roseus*, Corynebacterium sp., etc. Fungal genera such as Amorphoteca, Neosartorya, Talaromyces, Graphium, etc. and yeast genera, namely, Candida, Yarrowia, Pichia are also reported to for degradation of hydrocarbon [86].

The principle of aerobic degradation of hydrocarbons is shown in Fig. 4. [87,88]. The initial intracellular attack of organic pollutants is an oxidative process and catalyzed by oxygenases and peroxidases. Peripheral degradation pathways convert to different intermediates. Biosynthesis of cell biomass occurs from the central precursor metabolites and the growth of the cell biomass is continued.

Biodegradation of biodiesel by microorganisms is a complex process and there is very little information available on this aspect. This degradation process is normally known as cometabolic biodegradation [89]. Like petroleum diesel, biodiesel can also be biodegraded by bacteria, yeast and fungi [90]. However, biodiesel is biologically more active because of having FAMEs with hydrocarbon chains and two oxygen atoms. Various enzymes or bacteria can attack these FAME structures naturally. In the process of cometabolic degradation, FAMEs are oxidized at the position of beta carbon (2nd carbon that attaches to a functional group) and degraded to acetic acid and a fatty acid [91]. These fatty acids then breakdown to

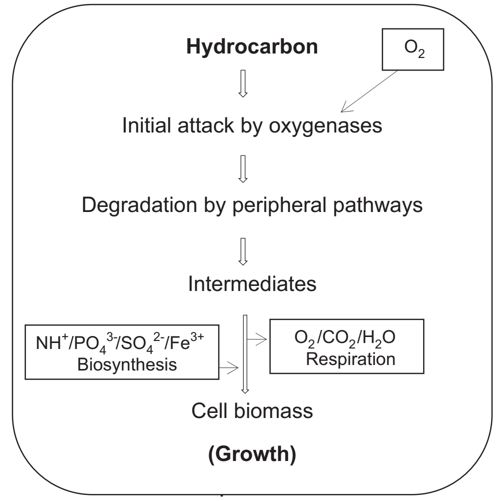



**Fig. 4.** Aerobic degradation of hydrocarbons by microorganisms (Adapted from Ref. [113,114]).

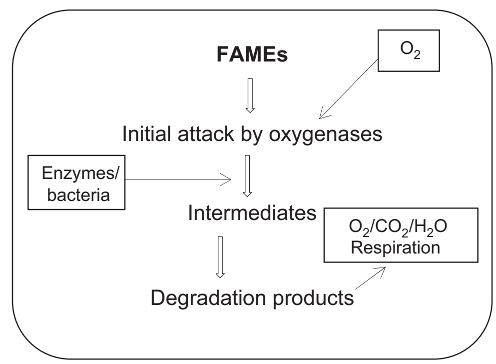



**Fig. 5.** Aerobic degradation of FAMEs by microorganisms (Adapted from Ref. [113]).

$CO_2$ and $H_2O$ by enzymes/bacteria by a series of reactions [92]. Simplified biodiesel biodegradation mechanism has been presented in Fig. 5 [92]:

Several studies reported that biodegradation of biodiesel and biodiesel blends occur more rapidly [84,89,93–96] than petroleum diesel. This could be attributed to the higher stability and chemical complexity of diesel than that of biodiesel. Diesel contains aliphatic cyclic hydrocarbons, benzene and so many other complicated hydrocarbons, which are very stable, and therefore, needs more energy for microorganisms to penetrate the structure[92]. On the other hand, biodiesel having oxygenated moieties can easily be subjected to attack by microorganisms. In addition, some inherent characteristics of biodiesel can enhance the degradation process. For example, storage conditions can facilitate the microbial growth to some extent. This statement was justified by investigations in aerobic and anaerobic storage conditions and results showed that anaerobic condition suppressed biodegradation more than aerobic conditions [96]. Due to hygroscopic nature of biodiesel, microbial growth occurs more rapidly in biodiesel and avoidance of water in the storage tanks are almost impossible to maintain. This residual water creates an interface to transport substrate (FAMEs) by enzymes that promote further degradation [97]. According to Demello et al. [75] both chemical corrosion and microbiologically influenced corrosion (MIC) can occur when water is present. In particular, problems with MIC may be enhanced by the use of biodiesel due to the higher bio-availability of this fuel compared to ordinary fossil fuel. De Beer and Stoodley [98] found that in fuel tanks containing oxygen, the conditions at the metal surface below a thin biofilm may be anaerobic and support growth of anaerobic

MIC-causing microorganisms. Joan Salvado et al. [95] found that density and viscosity of biodiesel blends deteriorated by biodegradation. Sorensen et al. [99] observed the presence of active anaerobic microorganisms in biodiesel blends after a certain period of experimentation.

Although several studies have reported the biodegradation of biodiesel by microbial growth, there are still adequate scopes of study on the effect of biodegradation on fuel properties and related mechanisms. At present, there is a lack of direct literatures on the effect of microbial growth showing the degradation of fuel properties such as oxidation stability, acid value, kinematic viscosity, cetane number, flash point, total contamination, calorific value, etc.

### 4.7. Combined effect of different factors

Different factors mentioned above do not affect the stability of biodiesel equally. Any specific property of biodiesel can be degraded by different factors. Inter-comparison with these factors has been summarized in Table 5.

From Table 5, it is evident that auto-oxidation degrades biodiesel's fuel properties more rapidly. Enhancement of oxidation rate occurs principally due to long time exposure and contact with metal surfaces. Almost all these factors change the chemical composition of biodiesel. In comparison to metal contamination and storage stability, microbial growth and water contamination were not so prominent to degrade fuel properties at every environment. The possible remedial measures could be (i) proper refining process during production, (ii) modification of storage conditions and (iii) addition of suitable additives. Further explanation on the possible remedial measures has been discussed in the following section.

## 5. Possible measures to improve stability

### 5.1. Refining during production

Biodiesel is generally produced through transesterification reaction [100]. The processing of biodiesel is greatly hinged on the rich scientific background and technology development for better process advancement [101]. To ensure the quality of biodiesel, raw materials first have to be processed. Some crude vegetable oils are associated with phospholipids and these phospholipids need to be removed by the hydration process [102]. Deodorization can be used effectively in order to eliminate free fatty acids, aldehydes, unsaturated hydrocarbons, and ketones, all of which cause undesirable odors and flavors of the oil [103]. High acid value of FFAs can be minimized by using iodine as a catalyst. Transesterification reaction is catalyzed by either homogeneous catalyst (sodium hydroxide, potassium hydroxide, sulfuric acid, hydrochloric acid, etc.) or heterogeneous catalyst (enzymes, alkaline earth metal compounds, titanium silicates, anion exchange resins, guanidine heterogenized on organic polymers, etc.). The alkali homogeneous catalysts are highly hygroscopic and absorb water from air during storage [104]. Therefore, they should be properly handled.

The alcohol materials used in the transesterification process normally include methanol, ethanol, propanol, butanol, and amyl alcohol. Once the transesterification reaction is completed, glycerol and alcohol are removed from the desired product esters by water washing [103,105]. Recently, the application of membrane technology for the separation and purification of biodiesel has tremendously reduced water washing application and has saved a reasonable amount of time and energy consumption [43]. Cooke et al. [106] practiced ion exchange resin to remove impurities. According to Gabelman and Hwang [107] hollow fiber membrane extraction can be an effective way to remove contaminants. These approaches effectively avoid loss of biodiesel yield during the production step and can improve the properties of fuel.

### 5.2. Addition of different additives

The stability of biodiesel in the presence of additives has been studied in details by several researchers [72,108,109]. Sarin et al. [110] have reported that two types of antioxidants are generally known: chain breakers and hydroperoxide decomposers. The two most common types of chain breaking antioxidants are phenolic and amine-types. Normally antioxidants act to inhibit the oxidation process and are well established for the use to control oxidation of biodiesel. For example, antioxidant (AH) intercepts the peroxide radical ($RCOO^{\bullet}$) in order to prevent it from creating another radical by the autoxidation mechanism. The related mechanism has been shown below [111]:

$$\text{Radical tapping stage}: \quad R-COO^{\bullet}+AH=R-COOH+A^{\bullet}$$
$$\text{Radical termination stage}: \quad A^{\bullet}+A^{\bullet}=A-A \text{ or non-radical materials}$$

Almost all the work related to stability of fatty oil and ester applications is limited to the phenolic type of antioxidants. The general structures of mostly used phenolic antioxidants are shown in Fig. 6.

The effectiveness of phenolic antioxidants depends on the number of hydroxyl/phenolic groups attached to its ortho, pera positions i.e. 1 and 2 or 1 and 4 positions in an aromatic ring. Generally, the active hydroxyl group can provide protons that inhibit the formation of free radicals or interrupt the propagation of free radical. Thus, it can delay the rate of oxidation. The effectiveness of TBHQ, PG, and PY can be explained based on their molecular structure. These additives possess two OH groups attached to the aromatic ring, while both BHT and BHA possess one OH group attached to the aromatic ring. Thus, on the basis of their electro-negativities, TBHQ, PG, and PY offer more sites for the

**Table 5**
Inter-comparison between different factors affecting the stability of biodiesel.

| Degradation criterion | Affecting factors | Remedial Measures |
|---|---|---|
| Induction period | Storage stability, thermal decomposition, metal contamination, auto-oxidation | Use of proper additives and modification of storage condition |
| Acid value | Auto-oxidation, storage stability, metal contamination | Use of proper additives |
| Iodine value | Auto-oxidation, storage stability, thermal decomposition, metal contamination | Use of proper additives and prohibition chemical degradation |
| Peroxide value | Storage stability, thermal decomposition, metal contamination, auto-oxidation | Using suitable additives |
| Viscosity | Auto-oxidation, storage stability, thermal decomposition, metal contamination, water absorption | Prevent chemical decomposition and sedimentation. |
| Density | Auto-oxidation, storage stability, thermal decomposition, metal contamination, water absorption, microbial growth, auto-oxidation | Prevent chemical decomposition andwater absorption. |
| Fatty acid composition | Auto-oxidation, storage stability, thermal decomposition, metal contamination, water absorption, microbial growth, auto-oxidation | Using proper refining process and modification of storage conditions |

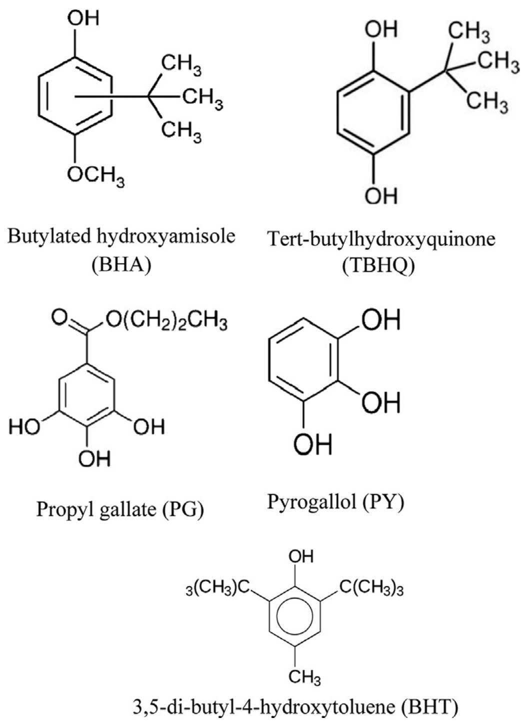



**Fig. 6.** Structure of different phenolic antioxidants [138].

**Table 6**
Relative effectiveness of anti-oxidants (Aos) for different biodiesels.

| Types of biodiesel | Relative effectiveness of antioxidants | Ref. |
|---|---|---|
| Soybean | TBHQ | |
| Jatropha | TBHQ > BHT > TBP > OBPA > a-T | |
| Palm biodiesel | TBHQ > BHT > BHA > a-tocopherol | |
| Waste frying oil | PG > BHA > BHT > TBHQ > vitamin E | |
| Soybean | Ferulic acid > callefic acid | [66] |
| Castor oil | BHA > PG > DBPC > TBHQ | |
| Sunflower | Callefic acid > chlorogenic acid | |
| Coconut oil | Callefic acid | |
| Rapeesed oil | PY > PG > TBHQ > BHT | |
| Sunflower oil ME (distilled) | TBHQ > PG > BHA > PY > BHT | |
| Used palm oil ME | PG > BHA > TBHQ > BHT > vitamin E | |
| Methyl soyate | BHA_PrG > TBHQ_BHT > a-tocopherol | |

formation of the complex between the free radical and antioxidant radical for the stabilization of the ester chain [112].

Antioxidants are used for delaying oxidation. From the literature reviews, it is evident that different antioxidants have different influences to improve the stability of different biodiesels. The relative effectiveness of antioxidants on different biodiesels is listed in Table 6.

From Table 6, it is seen that TBHQ is the most effective antioxidant for soybean, jatropha, palm and sunflower biodiesel. BHT, PG, and BHA are also found to be effective for almost all sort of biodiesels. It has been reported that use of several antioxidants can enhance the fuel stability up to the standard specification. The concentration of these additives may vary for different biodiesel and biodiesel blends because of the unsaturation concentration and composition. There are still many possible scopes of study in order to understand the effectiveness of different additives in enhancing corrosion resistance, stability as well as fuel properties of different biodiesels. This may contribute in using biodiesel at larger commercial scale.

### 5.3. Modification of storage conditions

There are several key factors that need to be considered for the storage of biodiesel, including oxidative stability, fuel solvency, and material compatibility [113,114]. Lee et al. [115] suggested that the storage temperature of most pure biodiesel need to be kept between 7 and 10 °C for better protection against oxidation. Underground storage of pure biodiesel ensures the storage temperature necessary for preventing crystal formation during extreme cold weather [116]. For more than 6 months storage pure biodiesel and its blends need to be treated with a suitable antioxidant. Biocides should be used because contamination from water may lead to biological growth in the fuel. Tanks for biodiesels storage should be made of Teflon, aluminum, fluorinated polyethylene or polypropylene and steel. ASTM D6751, EN 14214 [76,117] are the general guidelines for maintaining fuel properties. In conclusion, it can be observed that temperature and air exposure are major important factors affecting the degradation of biodiesel. If biodiesel is stored in high temperature environment and allowed to expose to ambient air, the degradation rate will be greatly increased. The temperature or air exposure alone, however, had little effect on biodiesel degradation. Water content in biodiesel will enhance biodiesel degradation due to hydrolysis but its effect is much less than other factors. Attention should therefore be paid to the above factors, particularly for long term storage of biodiesel [114]. The tanks should minimize the possibility of water contamination and should be cleaned prior to use for biodiesel storage [49].

## 6. Summary and conclusions

The properties of biodiesel could be affected by seven major factors, including (i) oxidation, (ii) thermal decomposition or thermal fluctuations, (iii) water absorption, (iv) biodegradation or microbial growth, (v) exposure to sunlight/photo oxidation, (vi) metal contamination and (vii) presence and absence of additives, etc. Although the properties of as-received biodiesel meet the given standard, many properties can deviate from the standard once biodiesel is degraded. As compared to petroleum diesel, biodiesel is thermally more instable. From the literature findings, it is found that temperature has a great influence in changing composition as well as other fuel properties. This could vary depending on the types of biodiesel and its compositional or molecular structures. Exposure of different metals can also influence the stability of biodiesel properties at different level. It was observed that due to metal exposure, induction period is reduced while acid value (AV), peroxide value (PV), and water content are increased. Degradation of these properties has been found to a greater extent due to exposure of copper as compared to other metals. Poor stability linked to different metal exposure could be another limitation for the commercial use of biodiesel.

In addition, biodiesel having oxygenated and unsaturated molecules are comparatively more subjected to microbial-degradation as compared to that of petroleum diesel. Although several studies have reported the biodegradation of biodiesel by microbial growth, there are still adequate scopes of study on related mechanisms and its effect on different fuel properties including oxidation stability, acid value, kinematic viscosity, cetane number, flash point, total contamination, calorific value, etc. Effect of different bacteria (e.g. *Pseudomonas fluorescens*, Bacillus sp., *Bacillus subtilis*, Alcaligenes sp., *P. aeruginosa*, *Acinetobacter lwoffi*, Flavobacterium sp., *Micrococcus roseus*, Corynebacterium sp., etc.), fungi (e.g. Amorphoteca, Neosartorya, Talaromyces, Graphium, etc.), yeasts (e.g. namely, Candida, Yarrowia, Pichia, etc.) on the degradation of biodiesel should be investigated.

A good number of works have been done so far on the stability of biodiesel but still there lies ample scope for further experimentations to minimize the effect of mentioned factors. Few additives (e.g. TBHQ, BHT, PG, BHA) have been reported to improve oxidation stability and few are (e.g. TBA, nBA, etc.) to reduce

corrosiveness. Some other additives may contribute in enhancing other fuel properties. Now it is important to develop such additives, which may ensure all these benefits. Recently, automobile industries have introduced a number of new materials for efficient fuel output and those materials have not yet been tested for biodiesel compatibility studies. In fact, still there is also a lack of scientific knowledge on the possible mechanisms of interaction of different automotive materials with biodiesel. Even a small improvement in material compatibility and stability of biodiesel may contribute towards the use of biodiesel comparatively at larger commercial scale.

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## References

[1] Lin L, Cunshan Z, Vittayapadung S, Xiangqian S, Mingdong D. Opportunities and challenges for biodiesel fuel. Applied Energy 2011;88:1020–31.
[2] Rizwanul Fattah I, Masjuki H, Liaquat A, Ramli R, Kalam M, Riazuddin V. Impact of various biodiesel fuels obtained from edible and non-edible oils on engine exhaust gas and noise emissions. Renewable and Sustainable Energy Reviews 2013;18:552–67.
[3] Meher L, Vidya Sagar D, Naik S. Technical aspects of biodiesel production by transesterification—a review. Renewable and Sustainable Energy Reviews 2006;10:248–68.
[4] Dorado M, Ballesteros E, Arnal J, Gomez J, Lopez F. Exhaust emissions from a diesel engine fueled with transesterified waste olive oil. Fuel 2003;82:1311–5.
[5] Khan SA, Hussain MZ, Prasad S, Banerjee U. Prospects of biodiesel production from microalgae in India. Renewable and Sustainable Energy Reviews 2009;13:2361–72.
[6] Silitonga AS, Masjuki HH, Mahlia TMI, Ong HC, Chong WT, Boosroh MH. Overview properties of biodiesel diesel blends from edible and non-edible feedstock. Renewable and Sustainable Energy Reviews 2013;22:346–60.
[7] Sendzikiene E, Makareviciene V, Janulis P. Influence of fuel oxygen content on diesel engine exhaust emissions. Renewable Energy 2006;31:2505–12.
[8] Demirbas A. Biodiesel: a realistic fuel alternative for diesel engines. Springer; 2008.
[9] Shahabuddin M, Liaquat A, Masjuki H, Kalam M, Mofijur M. Ignition delay, combustion and emission characteristics of diesel engine fueled with biodiesel. Renewable and Sustainable Energy Reviews 2013;21:623–32.
[10] Fazal M, Haseeb A, Masjuki H. Biodiesel feasibility study: an evaluation of material compatibility; performance; emission and engine durability. Renewable and Sustainable Energy Reviews 2011;15:1314–24.
[11] Demirbas A. Biodiesel production via non-catalytic SCF method and biodiesel fuel characteristics. Energy Conversion and Management 2006;47:2271–82.
[12] Aquino I, Hernandez R, Chicoma D, Pinto H, Aoki I. Influence of light, temperature and metallic ions on biodiesel degradation and corrosiveness to copper and brass. Fuel 2012;102:795–807.
[13] Haseeb ASMA, Masjuki HH, Ann LJ, Fazal MA. Corrosion characteristics of copper and leaded bronze in palm biodiesel. Fuel Processing Technology 2010;91:329–34.
[14] Haseeb A, Sia S, Fazal M, Masjuki H. Effect of temperature on tribological properties of palm biodiesel. Energy 2010;35:1460–4.
[15] Fazal M, Haseeb A, Masjuki H. Effect of temperature on the corrosion behavior of mild steel upon exposure to palm biodiesel. Energy 2011;36:3328–34.
[16] Haseeb A, Fazal M, Jahirul M, Masjuki H. Compatibility of automotive materials in biodiesel: a review. Fuel 2011;90:922–31.
[17] Fazal M, Haseeb A, Masjuki H. Corrosion mechanism of copper in palm biodiesel. Corrosion Science 2013;67:50–9.
[18] Fazal M, Haseeb A, Masjuki H. Degradation of automotive materials in palm biodiesel. Energy 2012;40:76–83.
[19] Fazal M, Haseeb A, Masjuki H. Comparative corrosive characteristics of petroleum diesel and palm biodiesel for automotive materials. Fuel Processing Technology 2010;91:1308–15.
[20] Singh B, Korstad J, Sharma Y. A critical review on corrosion of compression ignition (CI) engine parts by biodiesel and biodiesel blends and its inhibition. Renewable and Sustainable Energy Reviews 2012;16:3401–8.
[21] Fazal M, Haseeb A, Masjuki H. Effect of different corrosion inhibitors on the corrosion of cast iron in palm biodiesel. Fuel Processing Technology 2011;92:2154–9.
[22] Knothe G. Dependence of biodiesel fuel properties on the structure of fatty acid alkyl esters. Fuel Processing Technology 2005;86:1059–70.
[23] Fazal M, Haseeb A, Masjuki H. Investigation of friction and wear characteristics of palm biodiesel. Energy Conversion and Management 2013;67:251–6.
[24] Dunn RO. Antioxidants for improving storage stability of biodiesel. Biofuels, Bioproducts and Biorefining 2008;2:304–18.
[25] Atabani A, Silitonga A, Badruddin IA, Mahlia T, Masjuki H, Mekhilef S. A comprehensive review on biodiesel as an alternative energy resource and its characteristics. Renewable and Sustainable Energy Reviews 2012;16:2070–93.
[26] Valente OS, Da Silva MJ, Pasa VMD, Belchior CRP, Sodré JR. Fuel consumption and emissions from a diesel power generator fuelled with castor oil and soybean biodiesel. Fuel 2010;89:3637–42.
[27] He C, Ge Y, Tan J, You K, Han X, Wang J. Characteristics of polycyclic aromatic hydrocarbons emissions of diesel engine fueled with biodiesel and diesel. Fuel 2010;89:2040–6.
[28] Anastopoulos G, Lois E, Karonis D, Kalligeros S, Zannikos F. Impact of oxygen and nitrogen compounds on the lubrication properties of low sulfur diesel fuels. Energy 2005;30:415–26.
[29] Kousoulidou M, Fontaras G, Ntziachristos L, Samaras Z. Biodiesel blend effects on common-rail diesel combustion and emissions. Fuel 2010;89:3442–9.
[30] Jin F, Zeng X, Cao J, Kawasaki K, Kishita A, Tohji K, et al. Partial hydrothermal oxidation of unsaturated high molecular weight carboxylic acids for enhancing the cold flow properties of biodiesel fuel. Fuel 2010;89:2448–54.
[31] Mamat R, Abdullah NR, Xu H, Wyszynski M, Tsolakis A. Effect of fuel temperature on performance and emissions of a common rail diesel engine operating with rapeseed methyl ester (RME). SAE technical paper. 2009:01-1896.
[32] Goodrum JW, Geller DP. Influence of fatty acid methyl esters from hydroxylated vegetable oils on diesel fuel lubricity. Bioresource Technology 2005;96:851–5.
[33] Holser RA, Harry-O'Kuru R. Transesterified milkweed seed oil as a biodiesel fuel. Fuel 2006;85:2106–10.
[34] Ilham Z, Saka S. Two-step supercritical dimethyl carbonate method for biodiesel production from *Jatropha curcas* oil. Bioresource Technology 2010;101:2735–40.
[35] Kaul S, Saxena R, Kumar A, Negi M, Bhatnagar A, Goyal H, et al. Corrosion behavior of biodiesel from seed oils of Indian origin on diesel engine parts. Fuel Processing Technology 2007;88:303–7.
[36] Benjumea P, Agudelo J, Agudelo A. Basic properties of palm oil biodiesel–diesel blends. Fuel 2008;87:2069–75.
[37] Tan KT, Lee KT, Mohamed AR. A glycerol-free process to produce biodiesel by supercritical methyl acetate technology: an optimization study via response surface methodology. Bioresource Technology 2010;101:965–9.
[38] Boey P-L, Maniam GP, Hamid SA. Biodiesel production via transesterification of palm olein using waste mud crab shell as a heterogeneous catalyst. Bioresource Technology 2009;100:6362–8.
[39] Rashid U, Anwar F, Moser BR, Knothe G. Moringa oleifera oil: a possible source of biodiesel. Bioresource Technology 2008;99:8175–9.
[40] Steinbach A, Loyall U, ZUMBRÄGEL B, HAIDER C, Spinnler G, Schlink R, et al. Quality control of biofuels. Petroleum Technology Quarterly 2008.
[41] Schober S, Mittelbach M. Influence of diesel particulate filter additives on biodiesel quality. European Journal of Lipid Science and Technology 2005;107:268–71.
[42] Luque R, Herrero-Davila L, Campelo JM, Clark JH, Hidalgo JM, Luna D, et al. Biofuels: a technological perspective. Energy & Environmental Science 2008;1:542–64.
[43] Atadashi I, Aroua M, Aziz AA. High quality biodiesel and its diesel engine application: a review. Renewable and Sustainable Energy Reviews 2010;14:1999–2008.
[44] Jain S, Sharma M. Stability of biodiesel and its blends: a review. Renewable and Sustainable Energy Reviews 2010;14:667–78.
[45] Church DF, Pryor WA. Free-radical chemistry of cigarette smoke and its toxicological implications. Environmental Health Perspectives 1985;64:111.
[46] Haseeb A, Masjuki H, Ann L, Fazal M. Corrosion characteristics of copper and leaded bronze in palm biodiesel. Fuel Processing Technology 2010;91:329–34.
[47] Monyem A, Van Gerpen J H. The effect of biodiesel oxidation on engine performance and emissions. Biomass and Bioenergy 2001;20:317–25.
[48] Tang H, Abunasser N, Wang A, Clark BR, Wadumesthrige K, Zeng S, et al. Quality survey of biodiesel blends sold at retail stations. Fuel 2008;87:2951–5.
[49] Mittelbach M, Gangl S. Long storage stability of biodiesel made from rapeseed and used frying oil. Journal of the American Oil Chemists' Society 2001;78:573–7.
[50] Yang Z, Hollebone BP, Wang Z, Yang C. Landriault M. Effect of storage period on the dominant weathering processes of biodiesel and its blends with diesel in ambient conditions. Fuel 2013;104:342–50.
[51] Bondioli P, Gasparoli A, Della Bella L, Tagliabue S, Toso G. Biodiesel stability under commercial storage conditions over one year. European Journal of Lipid Science and Technology 2003;105:735–41.
[52] Leung DYC, Koo BCP, Guo Y. Degradation of biodiesel under different storage conditions. Bioresource Technology 2006;97:250–6.
[53] Joyner N, McIntyre J. The oven test as an index of keeping quality. Oil & Soap 1938;15:184–6.
[54] Sonntag N. Composition and characteristics of individual fats and oils. Bailey's Industrial Oil and Fat Products 1979;1:289–477.
[55] Joyner N, McIntyre J. The oven test as an index of keeping quality. Journal of the American Oil Chemists' Society 1938;15:184–6.

[56] Johnson OC, Kummerow F. Chemical changes which take place in an edible oil during thermal oxidation. Journal of the American Oil Chemists' Society 1957;34:407–9.
[57] Wexler H. Polymerization of drying oils. Chemical Reviews 1964;64:591–611.
[58] Dos Santos VML, Da Silva JAB, Stragevitch L, Longo RL. Thermochemistry of biodiesel oxidation reactions: a DFT study. Fuel 2011;90:811–7.
[59] Horel AA. Biodegradation of petroleum and alternative fuel hydrocarbons in moderate to cold climate: University of Alaska Fairbanks; 2009.
[60] Xin J, Imahara H, Saka S. Oxidation stability of biodiesel fuel as prepared by supercritical methanol. Fuel 2008;87:1807–13.
[61] Haseeb ASMA, Sia SY, Fazal MA, Masjuki HH. Effect of temperature on tribological properties of palm biodiesel. Energy 2010;35:1460–4.
[62] Haşimoğlu C, Ciniviz M, Özsert İ, İçingür Y, Parlak A, Sahir Salman M. Performance characteristics of a low heat rejection diesel engine operating with biodiesel. Renewable Energy 2008;33:1709–15.
[63] Lapuerta M, Rodríguez-Fernández J, Ramos Á, Álvarez B. Effect of the test temperature and anti-oxidant addition on the oxidation stability of commercial biodiesel fuels. Fuel 2012;93:391–6.
[64] Nzikou J, Matos L, Moussounga J, Ndangui C, Pambou-Tobi N, Bandzouzi E, et al. Study of oxidative and thermal stability of vegetable oils during frying. Research Journal of Applied Sciences 2009;4:94–100.
[65] Fernandes DM, Serqueira DS, Portela FM, Assunção RMN, Munoz RAA, Terrones MGH. Preparation and characterization of methylic and ethylic biodiesel from cottonseed oil and effect of tert-butylhydroquinone on its oxidative stability. Fuel 2012;97:658–61.
[66] dos Reis Albuquerque A, Maul J, dos Santos JP, dos Santos IMG, de Souza AG. Estimation of the oxidation temperature of biodiesels from a limited number of chemical parameters. Fuel 2012;102:585–91.
[67] Garcia C, Franco PI, Zuppa T, Antoniosi Filho N, Leles MI. Thermal stability studies of some cerrado plant oils. Journal of Thermal Analysis and Calorimetry 2007;87:645–8.
[68] Dunn RO. Effect of oxidation under accelerated conditions on fuel properties of methyl soyate (biodiesel). Journal of the American Oil Chemists' Society 2002;79:915–20.
[69] Sarin A, Arora R, Singh N, Sharma M, Malhotra R. Influence of metal contaminants on oxidation stability of Jatropha biodiesel. Energy 2009; 34:1271–5.
[70] Tagliabue S, Gasparoli A, Della Bella L, Bondioli P. Influenza della contaminazione metallica sulla stabilità termico ossidativa del biodiesel. Rivista Italiana delle Sostanze Grasse 2005;82:93–6.
[71] Wang Y-F, Huang K-L, Li C-T, Mi H-H, Luo J-H, Tsai P-J. Emissions of fuel metals content from a diesel vehicle engine. Atmospheric Environment 2003;37:4637–43.
[72] McCormick R, Ratcliff M, Moens L, Lawrence R. Several factors affecting the stability of biodiesel in standard accelerated tests. Fuel Processing Technology 2007;88:651–7.
[73] Fazal MA, Haseeb ASMA, Masjuki HH. Effect of temperature on the corrosion behavior of mild steel upon exposure to palm biodiesel. Energy 2011; 36:3328–34.
[74] Jain S, Sharma MP. Correlation development between the oxidation and thermal stability of biodiesel. Fuel 2012;102:354–8.
[75] DeMello JA, Carmichael CA, Peacock EE, Nelson RK, Samuel Arey J, Reddy CM. Biodegradation and environmental behavior of biodiesel mixtures in the sea: an initial study. Marine Pollution Bulletin 2007;54:894–904.
[76] Burton R An overview of ASTM D6751: biodiesel standards and testing methods. Alternative Fuels Consortium; 2008.
[77] Demirbas A. Progress and recent trends in biodiesel fuels. Energy Conversion and Management 2009;50:14–34.
[78] Barnes C, Garwood D, Price T. The use of biodiesel blends in domestic vaporising oil burners. Energy 2010;35:501–5.
[79] Srivastava A, Prasad R. Triglycerides-based diesel fuels. Renewable and Sustainable Energy Reviews 2000;4:111–33.
[80] Felizardo P, Neiva Correia MJ, Raposo I, Mendes JF, Berkemeier R, Bordado JM. Production of biodiesel from waste frying oils. Waste Management 2006; 26:487–94.
[81] Crookes R. Comparative bio-fuel performance in internal combustion engines. Biomass and Bioenergy 2006;30:461–8.
[82] Sharma Y, Singh B, Upadhyay S. Advancements in development and characterization of biodiesel: a review. Fuel 2008;87:2355–73.
[83] Eggins H, Oxley T. Biodeterioration and biodegradation. International Biodeterioration & Biodegradation 2001;48:12–5.
[84] Makareviciene V, Janulis P. Environmental effect of rapeseed oil ethyl ester. Renewable Energy 2003;28:2395–403.
[85] Adebusoye SA, Ilori MO, Amund OO, Teniola OD, Olatope S. Microbial degradation of petroleum hydrocarbons in a polluted tropical stream. World Journal of Microbiology and Biotechnology 2007;23:1149–59.
[86] Daugulis AJ, McCracken CM. Microbial degradation of high and low molecular weight polyaromatic hydrocarbons in a two-phase partitioning bioreactor by two strains of Sphingomonas sp. Biotechnology Letters 2003;25:1441–4.
[87] Fritsche W, Hofrichter M. Aerobic degradation by microorganisms. Biotechnology 2000;11:146–64.
[88] Das N, Chandran P. Microbial degradation of petroleum hydrocarbon contaminants: an overview. Biotechnology Research International 2011;2011:1–13.
[89] Zhang X, Peterson C, Reece D, Haws R, Moller G. Biodegradability of biodiesel in the aquatic environment. Transactions of the ASAE 1998;41:1423–30.
[90] Sorensen G, Pedersen DV, Norgaard AK, Sorensen KB, Nygaard SD. Microbial growth studies in biodiesel blends. Bioresource Technology 2011;102: 5259–64.
[91] Zubay G. Biochemistry. Wm. C. Brown Publishers, Dubuque. 1993;302312223:2.
[92] Cole GM. Assessment and remediation of petroleum contaminated soils: CRC Press; 1994.
[93] Prince RC, Haitmanek C, Lee CC. The primary aerobic biodegradation of biodiesel B20. Chemosphere 2008;71:1446–51.
[94] Owsianiak M, Chrzanowski Ł, Szulc A, Staniewski J, Olszanowski A, Olejnik-Schmidt AK, et al. Biodegradation of diesel/biodiesel blends by a consortium of hydrocarbon degraders: effect of the type of blend and the addition of biosurfactants. Bioresource Technology 2009;100:1497–500.
[95] Pasqualino JC, Montané D, Salvadó J. Synergic effects of biodiesel in the biodegradability of fossil-derived fuels. Biomass and Bioenergy 2006;30:874–9.
[96] Schleicher T, Werkmeister R, Russ W, Meyer-Pittroff R. Microbiological stability of biodiesel–diesel-mixtures. Bioresource Technology 2009;100:724–30.
[97] von Wallbrunn A, Richnow HH, Neumann G, Meinhardt F, Heipieper HJ. Mechanism of cis-trans isomerization of unsaturated fatty acids in *Pseudomonas putida*. Journal of Bacteriology 2003;185:1730–3.
[98] Beer D, Stoodley P. Microbial biofilms. In: Rosenberg E, DeLong E, Lory S, Stackebrandt E, Thompson F, editors. The prokaryotes. Berlin Heidelberg: Springer; 2013. p. 343–72.
[99] Sørensen G, Pedersen DV, Nørgaard AK, Sørensen KB, Nygaard SD. Microbial growth studies in biodiesel blends. Bioresource Technology 2011;102:5259–64.
[100] Atadashi I, Aroua M, Abdul Aziz A, Sulaiman N. The effects of water on biodiesel production and refining technologies: a review. Renewable and Sustainable Energy Reviews 2012;16:3456–70.
[101] Oh PP, Lau HLN, Chen J, Chong MF, Choo YM. A review on conventional technologies and emerging process intensification (PI) methods for biodiesel production. Renewable and Sustainable Energy Reviews 2012;16:5131–45.
[102] Van Gerpen J, Dvorak B. The effect of phosphorus level on the total glycerol and reaction yield of biodiesel. Bioenergy 2002. In: Proceedings of the 10th biennial bioenergy conference, Boise, ID2002.
[103] Demirbaş A, Kara H. New options for conversion of vegetable oils to alternative fuels. Energy Sources, Part A 2006;28:619–26.
[104] Leung D, Guo Y. Transesterification of neat and used frying oil: optimization for biodiesel production. Fuel Processing Technology 2006;87:883–90.
[105] Chen Y-c, S-l HE, Cheng J. Study on extraction and deacidification of oils and fats of natural plants by supercritical carbon dioxide. Journal of Yangtze University (Natural Science Edition) Sci & Eng V 2007;1:015.
[106] Cooke BS, Abrams C, Bertram B. Purification of biodiesel with adsorbent materials. CA Patent CA2,541,327; 2005.
[107] Gabelman A, Hwang S-T. Hollow fiber membrane contactors. Journal of Membrane Science 1999;159:61–106.
[108] Xin J, Imahara H, Saka S. Kinetics on the oxidation of biodiesel stabilized with antioxidant. Fuel 2009;88:282–6.
[109] Knothe G. Some aspects of biodiesel oxidative stability. Fuel Processing Technology 2007;88:669–77.
[110] Sarin A, Arora R, Singh N, Sarin R, Malhotra R. Blends of biodiesels synthesized from non-edible and edible oils: influence on the OS (oxidation stability). Energy 2010;35:3449–53.
[111] Pullen J, Saeed K. An overview of biodiesel oxidation stability. Renewable and Sustainable Energy Reviews 2012;16:5924–50.
[112] Karavalakis G, Stournas S. Impact of antioxidant additives on the oxidation stability of diesel/biodiesel blends. Energy & Fuels 2010;24:3682–6.
[113] Mudge SM, Pereira G. Stimulating the biodegradation of crude oil with biodiesel preliminary results. Spill Science & Technology Bulletin 1999;5:353–5.
[114] Leung D, Koo B, Guo Y. Degradation of biodiesel under different storage conditions. Bioresource Technology 2006;97:250–6.
[115] Lee I, Johnson LA, Hammond EG. Use of branched-chain esters to reduce the crystallization temperature of biodiesel. Journal of the American Oil Chemists' Society 1995;72:1155–60.
[116] Van Gerpen J, Shanks B, Pruszko R, Clements D, Knothe G. Biodiesel production technology. Report for the National Renewable Energy Laboratory USA: Department of Energy. 2004;30:42.
[117] Mittelbach M. Diesel fuel derived from vegetable oils, VI: specifications and quality control of biodiesel. Bioresource Technology 19967–11.